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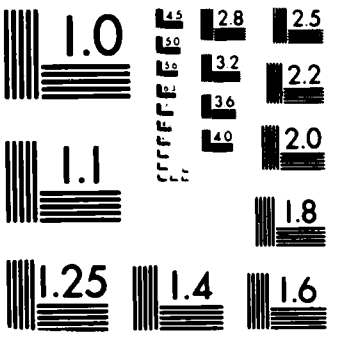
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The circadian rhythm in body temperature is thought to be due to a rhythm in the thermoregulatory "set-point". The overall goal of our research was to determine whether this represents a circadian "fever". If this hypothesis is correct, then antipyretic drugs should attenuate the rhythm in body temperature. We have found that administration of a variety of antipyretic drugs to rats markedly reduced their nighttime elevation in body temperature. These data suggest to us that prostaglandins are probably involved in the circadian rhythm in body temperature. We have attempted to determine whether this rhythm in prostaglandins is dependent on a rhythm in circulating concentrations of interleukin-1 (IL-1). Based on studies with rats and human beings using bioassays and immunoassays, we have been unable to detect any rhythm in plasma concentration of IL-1. We conclude that it is unlikely that circulating IL-1 has a role in the rhythm in body temperature. If IL-1 influences the circadian rhythm in body temperature, it may do so at the level of the hypothalamus or at some other central nervous site.

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The original specific aims in our grant application were:

1. to determine whether the circadian changes in body temperature, iron and zinc was attributable to a circadian variation in the plasma concentration of interleukin-1 (IL-1),
2. to determine whether the enhanced survival rate of rats to infection with Salmonella typhimurium at night was attributable to an increased production and release of IL-1 at night,
3. to determine whether the circadian changes in temperature, iron and zinc was due, in part, to a circadian pattern in some signal from the gastrointestinal tract, and
4. to determine whether there is a circadian pattern in the plasma concentration of IL-1 in human beings.

Three years of research support was requested; two years were granted. Substantial progress has been made towards answering specific aims 1, 3, and 4.

A biotelemetry system has been established that enables us to monitor and record temperature and activity in freely moving small animals. We have used this system for our studies on circadian rhythms in rats and mice. The nighttime rise in body temperature in rats can be blocked by intraperitoneal (ip) injection of the antipyretic drugs indomethacin, sodium salicylate and acetylsalicylic acid (Scales and Kluger, In Press, 1987). This reduction in nighttime temperature is not the result of decreased activity. Since antipyretic drugs are thought to block fevers by inhibiting the production of prostaglandins of the E series (PGE) in the hypothalamus, our data suggest to us that there is a circadian variation in PGE in this region of the central nervous system. In a related series of studies, we have attempted to determine whether stress-induced hyperthermia is a prostaglandin-mediated event, blockable by antipyretic drugs. Intraperitoneal injection of either sodium salicylate or indomethacin attenuated this stress-induced rise in body temperature (Singer et al., 1986; Kluger et al., 1987). We also found that intracerebroventricular administration of sodium salicylate blocked a significant portion of the rise in body temperature that results from exposure of rats to a novel environment. We are currently funded by the NIH to investigate whether IL-1 or other mediators of the acute phase response are responsible for a portion of these stress-induced rises in

body temperature.

It is generally accepted that fever, whether it be the result of infection with bacteria, viruses, or other pathogens, is attributable to the release of IL-1 into the circulation. The above data on circadian rhythms in body temperature are consistent with there being a circadian variation in plasma concentration of IL-1. To test this hypothesis, plasma was obtained from rats in the morning and in the evening. This plasma was column chromatographed (G-50) to remove a 40kd inhibitor that is found in plasma (Cannon and Dinarello, 1985), and then assayed in a sensitive two cell IL-1 assay (the LBRM33 1A5/HT2 assay) developed by Gillis and Mizel (1981). The basis for this assay is that the LBRM33 1A5 cells produce IL-2 in response to mitogen and IL-1. The supernatant from stimulated LBRM33 1A5 is then added to freshly plated HT2 cells, which are dependent on IL-2 for growth. Growth of HT2 cells is measured by incorporation of radiolabelled thymidine. This assay can measure IL-1 at concentrations considerably lower than 1 unit per ml of plasma. No circulating IL-1 was found in the plasma of rats either during the daytime or nighttime.

Based on the negative results obtained above, we ran several "positive" controls. In one study, IL-1 sufficient to produce a large fever was injected into rats. When this plasma was chromatographed, large amounts of IL-1 were recovered; however, when endotoxin was injected into other rats (also a concentration sufficient to produce a large fever), and this plasma was chromatographed, no IL-1 was recovered from the plasma. These data suggest to us that circulating IL-1 may not be the critical factor to measure during either endotoxin-induced fever or during circadian rhythms (manuscript in preparation). It is possible that IL-1 produced locally (e.g. in the central nervous system by glial cells) is responsible for a circadian cycle in brain PGE. In situ hybridization experiments are currently in progress to attempt to address this question.

A study was run in our Clinical Research Center in which we monitored body temperature, plasma iron and zinc concentration, and circulating white blood cell populations in 8 human subjects for two two-day sessions. As in our earlier study using rats, we found that there is an inverse rhythm between body temperature and plasma trace metals. To determine whether plasma concentration of IL-1 could be driving these rhythms, plasma samples were monitored for IL-1 using a radioimmunoassay developed by Cistron Technology (reported to be able to measure IL-1 to ca. 260 picograms) and an ELISA developed by Syntex

Corporation (reported to be able to measure IL-1 to ca. 15 picograms). The results of both assays indicated that there is no detectable rhythm in plasma concentration of IL-1 in human beings (Scales et al, manuscript in preparation).

To determine whether a signal from the gastrointestinal tract (e.g. endotoxin) could be influencing the rhythm in body temperature, rats were implanted with Alzet miniosmotic pumps containing polymyxin (a cationic antibiotic that inactivated some endotoxin) or saline (controls). Polymyxin did not affect daytime body temperature, but totally blocked the circadian rise in body temperature of rats for several days. These data suggested to us that endotoxin might be leaking from the gastrointestinal tract and that this leakage could influence the rhythm in body temperature. In a subsequent study, rats were given streptomycin/bacitracin (broad-spectrum nonabsorbable antibiotics) in their drinking water. This resulted in a reduction in both the daytime and nighttime body temperature without significantly affecting the rhythm in body temperature. The next question we attempted to address was whether germfree animals have a dampened amplitude in their body temperature rhythm. Germfree mice (CD-1) were implanted with biotelemeters and body temperature was recorded for several weeks. Compared to conventional mice, the amplitude in the rhythm in body temperature in germfree mice is markedly attenuated. In a preliminary study, we have found that "conventionalizing" germfree mice resulted in a rhythm in body temperature indistinguishable from control (conventional) CD-1 mice. These data suggest to us that the rhythm in body temperature in mice is dependent on multiple signals. The amplitude of the rhythm (i.e. the difference between the average daytime and average nighttime temperature) appears to be dependent to about 50% on signals not involving the presence of gut flora and to about 50% on signals originating from the intestinal flora. These data have formed the basis for a grant application designed to investigate the role of intestinal flora in circadian rhythms in body temperature ("Body Temperature Rhythms and Gut Flora") that is currently being reviewed by the NIH.

References Cited

Cannon, J.G., and C. A. Dinarello. Increased plasma interleukin-1 activity in women after ovulation. Science 227:1247-1249, 1985.

Gillis, S. and S.B. Mizel. T-cell lymphoma model for the analysis of interleukin-1 mediated T-cell activation. Proc Nat Acad Sci USA, 78:1133-1137, 1981.

Publications Resulting From This Research Support

Singer, R., Harker, C.T., Vander, A.J., and Kluger, M.J. Hyperthermia induced by open-field stress is blocked by salicylate. Physiol. Behav. 36:1179-1182, 1986.

Kluger, M.J., O'Reilly, B., Shope, T. R., and Vander, A.J. Further evidence that stress hyperthermia is a fever. Physiol. Behav. 39:763-766, 1987.

Scales, W.E., and Kluger, M.J. Antipyretic drugs attenuate the circadian rise in body temperature of the rat. Amer. J. Physiol., In Press, 1987.

Manuscripts In Preparation

Scales, W.E., Vander, A.J., and Kluger, M.J. Circadian variation in body temperature and trace metals in human beings.

Scales, W.E., O'Reilly, B., Vander, A.J., and Kluger, M.J. Evidence that circulating IL-1 is not involved in endotoxin-induced fever.

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